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SOIL STRUCTURE AND MYCORRHIZAE ENCOURAGE BLACK WALNUT GROWTH ON OLD FIELDS

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ABSTRACT.—Examination of black walnut seedlings grown in forest and field soils showed all root systems were infected with mycorrhizae; the amount of infection was influenced by treatments. Mean height and dry weight of tops and roots were greater for seedlings grown in forest than field soil. Seedling height growth was not increased by disturbing either soil; but, root dry weight was significantly increased by disturbing the field soil.

OXFORD: 114.14:181.351:176.1 *Juglans nigra*.
KEY WORDS: Endomycorrhizal fungi, bulk density sampling, soil sterilization.

Black walnut trees (*Juglans nigra* L.) planted on abandoned fields often grow much less than trees planted on cleared forest sites or on old fields reverted to tree cover (Carmean *et al.* 1976). Good soil structure, valuable for promoting plant growth and microorganisms, is less common in old fields than in the undisturbed soils under continuous forest cover.

Black walnut is known to form vesicular-arbuscular (VA) mycorrhizae. Mazur and Semahanova (1965) reported that mycorrhizae developed best on *J. regia* L. at 40 to 50 percent moisture content, and seedling development (foliation, height, stem diameter, and root system) was positively related to the amount of mycorrhizae formation.

This report describes the influence of disturbed and undisturbed soils from both forested and abandoned field sites, and the effects of inoculation with VA endomycorrhizal fungi on the growth of black walnut seedlings under greenhouse conditions.

METHODS

In the spring of 1978, soils of the Wellston series (fine-silty, mixed, mesic-Ultic Hapludalfs) were collected from a forest and an abandoned field on the Kaskaskia Experimental Forest in Hardin County, Illinois. This silt loam soil is well-drained and was formed in loess and underlying material weathered from sandstone or shale bedrock. Topsoil thickness averaged 12 cm in the forest and 8 cm in the abandoned field. Subsoil thickness averaged nearly 1 m to sandstone bedrock in both soils. The abandoned field (last cultivated in early 1960's) was dominated with broomsedge (*Andropogon virginicus* L.), and had been sparsely invaded by white ash (*Fraxinus americana* L.), green ash (*F. pennsylvanica* var. *subintegerrima* (Vahl.) Fern.), yellow-poplar (*Liriodendron tulipifera* L.), sassafras (*Sassafras albidum* Nutt.), and flowering dogwood (*Cornus florida* L.). The forest area is adjacent to the field and has sawtimber-size white oak (*Quercus alba* L.), white ash, yellow-poplar, and several large diameter black walnut (*Juglans nigra* L.) trees.

Using a newly designed soil sampler and cast acrylic resin tubes, relatively undisturbed soil cores 20 cm in diameter were obtained from the upper 36 cm of both forest and field. Sections of burlap were fastened over the bottom ends of the tubes to retain soil. Soils for the disturbed treatment were also collected from the upper 36 cm of the forest and field. This soil was thoroughly disturbed by the use of a mechanical shredder and equal amounts (by weight) were put into epoxy painted (inside and outside) galvanized steel containers the same size as the acrylic containers and secured with burlap.

Sixty containers each of the forest and field soils were prepared. Treatments included undisturbed (U), disturbed (D) undistrubed autoclaved (UA), disturbed autoclaved (DA), undisturbed, autoclaved, and inoculated (UAI), and disturbed, autoclaved, and inoculated (DAI). Each treatment was replicated 10 times for each of the two soils. All containers were randomly placed in a shaded greenhouse in mid-May 1978. The acrylic containers were covered with aluminum foil to prevent roots from being affected by sunlight.

Autoclaved soils were treated at 122C and pressure of 1.5 kg/cm^2 in containers for two 2-hour periods separated by 24 hours. Autoclaved and inoculated soils were inoculated by forcing a 10 mm sterilized glass tube into the soil to a depth of 10 cm in each container at three equi-distant locations, removing a plug of soil, and inserting a sterilized glass tube of nonautoclaved forest soil into the hole. An additional 10 g of nonautoclaved forest soil was broadcast over the surface to complete inoculation.

One pregerminated black walnut seed that had been surface-sterilized with sodium hypochlorite was planted in each container. Seedlings were watered with distilled water as needed throughout the growth period.

After 12 weeks of growth, seedlings were harvested and stem height and diameter (2.54 cm above root collar) were recorded. Roots were separated from tops and rinsed thoroughly in tap water. Six 10-cm lengths per treatment were randomly removed and examined microscopically at 100X for the presence of VA mycorrhizal infection after clearing and staining according to the method of Phillips and Hayman (1970). The root was scored mycorrhizal if hyphae, arbuscules, or vesicles were present separately or together in any part of the root segment. The density of the infection was not measured. The remaining roots and tops were ovendried for 24 hours at 65C and weighed.

RESULTS AND DISCUSSION

Mycorrhizae

Roots of seedlings grown in all treatments of the undisturbed and disturbed forested and field soils were mycorrhizal, but the amount of infection was influenced by treatments (figs. 1D and H). Autoclaving undisturbed forest soil cores (UA) reduced mycorrhizal infection to 40 percent, but autoclaving undisturbed field soil cores reduced mycorrhizal infection only to 80 percent. When undisturbed and autoclaved forest soil cores (UA) were inoculated (UAI) the mycorrhizal infection increased from 40 to 60 percent and in the case of the field soil (UAI), roots were 100 percent infected as were seedlings in the undisturbed field cores. Thus, it can be concluded that autoclaving undisturbed soils at 122C reduces mycorrhizal infection but does not completely eliminate infection.

Results were similar for seedlings grown in disturbed and autoclaved cores for both forest and field soils (fig. 1D and H). Autoclaving disturbed forest and field soils (DA) reduced mycorrhizal infection much more than autoclaving undisturbed soils (UA), and when these disturbed and autoclaved soils were inoculated (DAI) mycorrhizal infection was greatly improved. The percent infection increased from 20 to 100 in roots of seedlings grown in field soil cores but only from 20 to 60 for seedlings in forest soil cores. Thus, it can be concluded that inoculation of both forest and field soils that have been autoclaved improved mycorrhizal infection. Also, the inoculum and the method used for inoculation, although improving infection, did not usually result in the 100 percent infection observed in soils that were not autoclaved.

Seedling growth

Comparison of mean height and dry weight of roots and tops shows that seedlings grown in containers of forest soil were superior to those in the field soil (fig. 1A,B,C,E,F,G). Much of the greater height, and top and root dry weights of

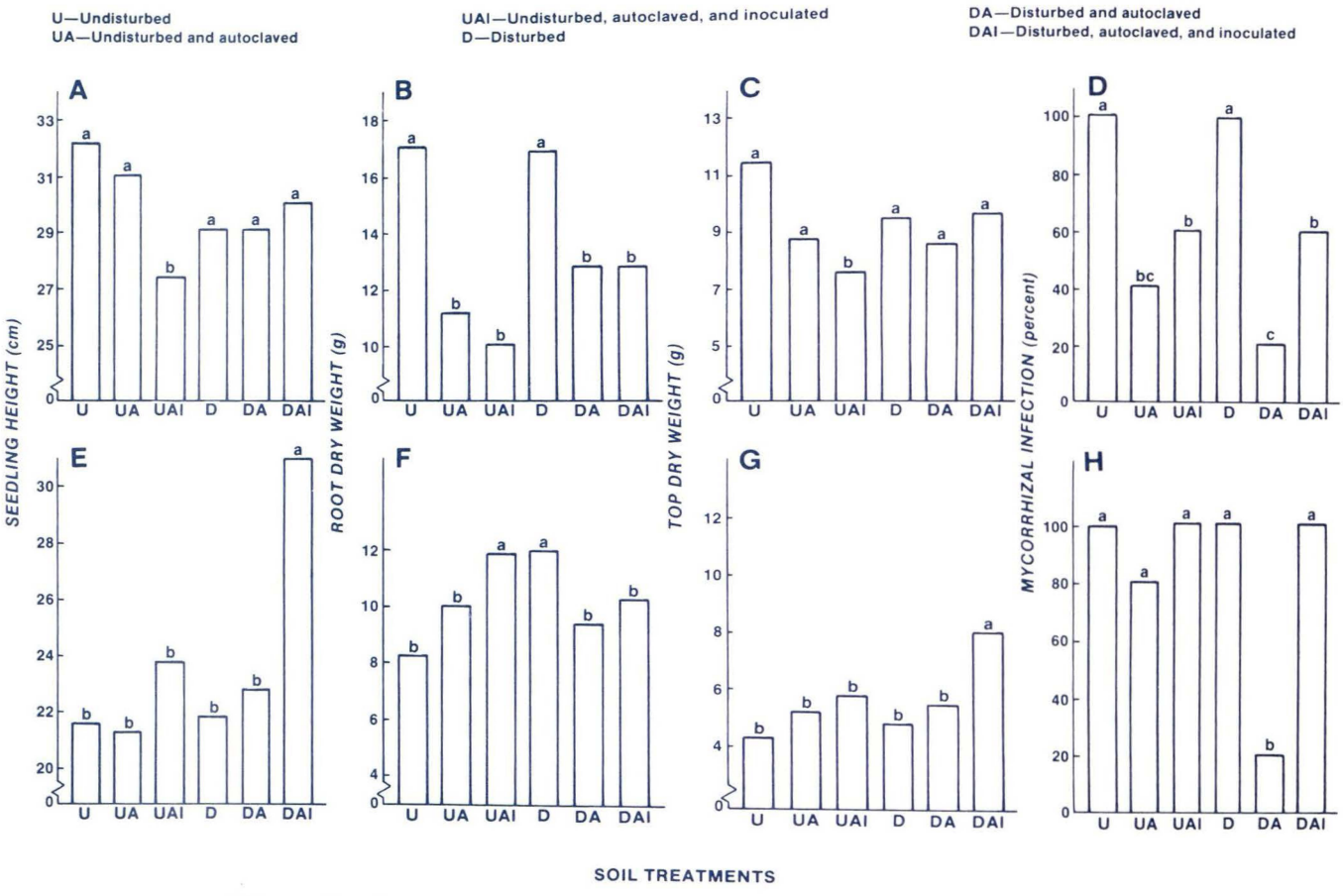

Figure 1.—*Effects of soil treatments on seedling height, root dry weight, top dry weight, and mycorrhizal infection in forest soils, A through D respectively; and field soils, E through H respectively. (Bars with the same letters are not significantly different at the 0.05 level. Means of 10 plants.)*

seedlings grown in forest soil may be attributed to its lower bulk density, and greater nutrient and organic matter content compared to field soil (table 1). Height growth of seedlings was not increased by disturbing the forest soil (D). Clark (1964) showed that yellow-poplar seedlings grown in undisturbed forest soil had greater height and dry weight than seedlings grown in disturbed forest soil.

The root dry weight of seedlings grown in disturbed field soil was significantly increased over seedlings grown in undisturbed field soil. Undisturbed field soils are dense (1.40 g/cm^3,table 1); disturbing these soils reduced bulk density to 1.18 g/cm^3. Thorough loosening of the soil may not always be desirable, as some plants require rather dense soil for best root-soil contact and growth. Evidently loosening the field soil in this study improved soil aeration and favored root penetration, proliferation, and growth. An increase in root mass in response to lowering the soil bulk density might lead to improved top growth differences in old field plantings years later.

Lack of mycorrhizae is likely not a problem in the growth of black walnut on abandoned field sites. However, the more dense, and less aerated condition of undisturbed field soils might have diminished VA mycorrhizal activity and thus reduced the ability of seedlings to absorb nutrients and moisture. We did not determine whether the mycorrhizae in forest and field soils are the same organisms. If they are different and some are more effective in promoting growth than others, infection alone may not mean much (Marx *et al.* 1971).

Table 1.—*Average soil properties before and after treatments*

Soil	pH[1]	Bulk density[2]	Organic matter[3]	Nutrients[4]				
				N	P	K	Ca	Mg
		g/cm³	*Percent*	*Ppm*				
Before								
Forest	6.1	1.29	.88	27.6	7.5	42	928	75
Field	5.3	1.40	0.94	3.6	5.4	33	395	120
After								
Forest	6.0	1.15	1.56	25.2	6.4	43	1404	75
Field	5.4	1.18	0.78	6.0	6.6	35	508	112

[1]pH by glass electrode in 1:1 soil solution.

[2]Bulk density according to the formula ovendry weight/volume of sample.

[3]Organic matter by titration after oxidation with 1N $K_2Cr_2O_7$ and concentrated H_2SO_4.

[4]N colorimetrically after extraction with $Ca(OH)_2$; P colorimetrically after extraction with 0.002N H_2SO_4; K, Ca, and Mg by atomic absorption after extraction with 0.075N acid mixture (0.5N HCl + 0.25N H_2SO_4).

Both broomsedge and sassafras in the abondoned field vegetation contain allelopathic compounds that have been known to inhibit the growth of several competing plant species (Rice 1972); others inhibit nitrogen fixing and nitrifying bacteria (Rice 1964, Gant and Clebsch 1975). Carmean *et al.* (1976) suggested that allelopathic compounds produced by broomsedge and sassafras may reduce the growth of planted hardwoods. In most cases reduced plant growth has been attributed to low soil nitrogen. There may be a connection between low soil nitrogen and allelopathy that is not yet completely understood (Rice 1977).

CONCLUSION

Black walnut seedlings grown in both forest and field soils were mycorrhizal to some degree. Forest soil had the best growth of all treatments, and seedlings were well infected with endomycorrhizae. Good seedling growth is apparently dependent on high mycorrhizal infection rates—rates that could not be achieved by autoclaving and then inoculating previously undisturbed forest soil. Disturbing field soils reduced bulk density and increased average root dry weight of seedlings. Even though establishment of mycorrhizal infection is no problem in old fields, soil structure may restrict optimal mycorrhizal development. We conclude that both mycorrhizae and soil structure are important in black walnut seedling growth.

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